AD-A227 829

DOCUMENTATION PAGE

Form Approved OMB No. 0704-0188

	1b RESTRICTIVE MARKINGS: NA
2a SECURITY CLASSIFICATION AUTHORITY: NA	3 DISTRIBUTION/AVAILABILITY OF REPORT: Distribution Unlimited
2b DECLASSIFICATION/DOWNGRADING SCHEDULE: NA	
4. PERFORMING ORGANIZATION REPORT NUMBER(S)	5. MONITORING ORGANIZATION REPORT NUMBER(S): NA
6a NAME OF PERFORMING ORGANIZATION: Hahnemann University; 6b OFFICE SYMBOL (If applicable): NA	7a. NAME OF MONITORING ORGANIZATION: Office of Naval Research
6c. ADDRESS (City, State, and ZIP Code): Broad and Vine, Philadelphia, PA 19102	7b. ADDRESS (City, State, and ZIP Code): 800 N. Quincy Street, Arlington, VA 22217-5000
8a NAME OF FUNDING/SPONSORING ORGANIZATION: Office of Naval Research; 8b OFFICE SYMBOL (If applicable): ONR	9 PROCUREMENT INSTRUMENT IDENTIFICATION NUMBER: N00014-90-J-1351
8c. ADDRESS (City, State, and ZIP Code): 800 N. Qunicy Street, Arlington, VA 22217-5000	10 SOURCE OF FUNDING NUMBERS: PROGRAM ELEMENT NO. / PROJECT NO. / TASK NO. 4414021 / WORK UNIT ACCESSION NO.

11 TITLE (Include Security Classification)
STRUCTURE AND FUNCTION OF POLYMERIZABLE PROTEIN/LIPID BILAYERS

12 PERSONAL AUTHOR(S)
Ahl, Patrick, Leonard

13a. TYPE OF REPORT	13b TIME COVERED	14. DATE OF REPORT (Year, Month, Day)	15 PAGE COUNT
Final	FROM 1-90 TO 10-90	1990 -9-14	3

16 SUPPLEMENTARY NOTATION
none

17 COSATI CODES			18 SUBJECT TERMS (Continue on reverse if necessary and identify by block number)
FIELD	GROUP	SUB-GROUP	membranes, polymerizable lipids
08			

19 ABSTRACT (Continue on reverse if necessary and identify by block number)

Title: Structure and Function of Polymerizable Protein/Lipid Bilayers

The long-range goal of this research was to develop "rugged" polymerized phospholipid bilayers suitable for biosensor applications. The specific objective was to examine how the addition of a non-polymerizable phosholipid effects the polymerization of diacetylenic phospholipid. Surprising, the addition of certain non-polymerizable lipid can enhance the polymerization of a particular diacetylenic lipid. This effect will be described in a paper by Ahl, et al. in Biochimica et Biophysica Acta which is in press.

20 DISTRIBUTION/AVAILABILITY OF ABSTRACT	21. ABSTRACT SECURITY CLASSIFICATION	
☒ UNCLASSIFIED/UNLIMITED ☐ SAME AS RPT ☐ DTIC USERS	(U)	
22a NAME OF RESPONSIBLE INDIVIDUAL: Dr. Igor Vodyanoy	22b TELEPHONE (Include Area Code): (202) 696-4506	22c OFFICE SYMBOL: ONR

DD Form 1473, JUN 86 — Previous editions are obsolete. — SECURITY CLASSIFICATION OF THIS PAGE

S/N 0102-LF-014-6603

90

Final Report

R&T Code: 4414021---01
September 14, 1990

FINAL REPORT ON CONTRACT N0014-90-J-1351

PRINCIPAL INVESTIGATOR: Dr. Patrick L. Ahl

CONTRACTOR: Hahnemann University

CONTRACT TITLE: Structure and Function of Polymerized Protein/Lipid Bilayers

START DATE: 1 January 1990

RESEARCH OBJECTIVE:
The long range goal was to develop "rugged" polymerized phospholipid bilayers suitable for biosensor applications. The specific objective of this project was to examine the how the addition a non-polymerizable phosphaditylcholines of various acyl chain lengths effects the polymerization of the diacetylenic phosphatidylchoine, 1,2-bis(tricosa-10,12-dinoyl)-*sn* - glycero-3-phosphocholine (DC8,9PC).

PROGRESS (8 MONTHS):
This project was proposed to cover a period of one year. I am terminating this project after 8 months because I am leaving Hahnemann University to take a research position with a biotechnology company which specializes in medical applications for liposomes. This report will summarize the progress made during this 8 month period.

During my tenure at the Naval Research Laboratory, Dr. Alok Singh and myself, developed a method to incorporate membrane proteins into polymerized bilayers composed of the polymerizable lipid DC89PC and the non-polymerizable lipid dinonanoyl-phosphatidylcholine (DNPC). We found that the short acyl chain phosphatidylcholine DNPC significantly improved the polymerization of DC89PC in the bilayer membrane. One of the principle goals of this project was to determine molecular basis for how a non-polymerizable lipid improves the polymerization of the diacetylenic lipid.

During this project, I examined what effect varying the acyl chain length of the non-polymerizable phosphatidylcholine had on the polymerization efficiency of DC89PC. The extent of polymerization was determined by UV/visible spectroscopy using a specially modified 4800 SLM fluorometer. The lipid samples were contained in a special variable pathlength , temperature controlled, CaF2 sample cell. The samples were polymerized in the sample cell at 4 C with UV irradiation from a 100 watt Hg lamp. High signal to noise UV/visible difference spectra were obtained with relatively small amounts of sample. For example, polymerization of 2:1 mole ratio DNPC/DC89PC membranes (10 mg/ml total lipid) produced absorbance changes of 0.6 to 0.7 a.u., even when the measuring light pathlength was only 100 microns. This extremely short pathlength CaF2 sample cell was used so that absorbance changes following polymerization could also be measured in the IR region of the spectrum. The ultimate goal was to examine the spectra changes induced by polymerization in both the UV/visible and IR regions. Although, preparations for IR measurements using a FTIR spectrometer were in progress, I was not able to make these measurements because of my departure from Hahnemann University.

The investigation of how the acyl chain length of the non-polymerizable lipid affect the polymerization of DC89PC using UV/visible spectroscopy had a somewhat surprising result. We already knew that phospholipid bilayers composed of 2:1 mole ratio DNPC-DC89PC clearly polymerize much more efficiently that bilayers containing only DC89PC. In addition, the polymerization efficiency of DC 89PC was also enhanced relative to pure DC89PC by addition of dimyristoylphosphaditylcholine (DMPC) or dipalmitoylphosphaditylcholine (DPPC), but to a much smaller degree. It was thought that the reduced effectiveness of DMPC and DPPC was due to the long acyl chains of these lipids, C14 and C16 respectively. The hypothesis was that the acyl chains would sterically interfere with the diacetylenic polymerization in the middle of the bilayer. If this hypothesis is true, then polymerization efficiency of DC89PC in combination with dilauralphosphatidylcholine DLPC (C12) should be better than with either DMPC or DPPC. However, contrary to what was expected addition of DLPC appeared to inhibit the polymerization of the DC89PC. The results suggest that tendency of DNPC to dramatically improve DC89PC polymerization is unique and that other factors in addition to acyl chain length maybe involved.

WORK PLAN (REMAINING 3 MONTHS):
Since I am leaving Hahnemann University to work in the biotechnology industry, I will not be continuing this ONR funded project.

PUBLICATIONS AND REPORTS (8 MONTHS):
During my tenure at Hahnemann I submitted a manuscript on my polymerizable lipid research done at the Naval Research Laboratory. The manuscript is entitled "Insertion of Bacteriorhodopsin into Polymerized Diacetylenic Phosphatidylcholine Bilayers" and was accepted for publication in Biochimica et Biophysica Acta.

TRAINING ACTIVITES:
None